THE HEREDITARY ANEMIAS*

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INTRODUCTION

THE anemias which have in common a hereditary factor are characterized by an intracorpuscular defect, and are hemolytic in type. With the exception of thalassemia the anemia is normocytic and normochromic. No extracorpuscular defect has been found to be present except where a complicating factor of "hypersplenism" has developed later on in the course of the disease. These important facts have been established by studies of the life span of the red blood cells. When the erythrocytes of the patient have been transfused into normal recipients, life spans of fourteen to sixty days, instead of the normal 120 days, have been found. This, then, constitutes an intracorpuscular defect. Conversely, normal erythrocytes transfused into these patients survive for 120 days, and thus the absence of an extracorpuscular defect is established.

Shortened survival of the patient's red blood cells implies rapid hemolysis within the body. This results in an increase of fecal and urinary urobilinogen, and of the serum bilirubin, largely of the "indirect" fraction. The bone marrow responds to the demand for more red blood cells by a heightened production, as is reflected in an increase of the reticulocytes. If the bone marrow is placed under unusually severe stress, normoblasts, and even increased numbers of leukocytes and platelets will appear in the peripheral blood. These evidences of increased blood destruction and production must always be looked for when one suspects a hemolytic anemia.

Many painstaking genealogic studies of the patients and their families have shown that in the case of congenital hemolytic anemia, ovalocytosis, and hereditary nonspherocytic hemolytic anemia, the disease is inherited as a Mendelian dominant from either parent. In the case of thalassemia and sicklemia homozygosity of the abnormal gene results in the expres-

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TABLE I—HEREDITARY ANEMIAS (INTRA CORPUSCULAR DEFECT)

I.	Congenital Hemolytic Anemia
II.	Thalassemia Major and Minor
III.	Sickle Cell Anemia
IV.	HGB. C and HGB. D Disease
V.	Combination
	(1) Sickle Cell—Thalassemia
	(2) Sickle Cell—HGB. C
	(3) Sickle Cell—HGB. D
VI.	Hereditary Non-spherocytic Hemolytic Anemia
VII.	Ovalocytosis

sion of the disease, and heterozygosity, the trait. It is not surprising, in view of the fact that the defect lies within the red blood cell, that this defect has been shown to be due to an abnormal hemoglobin in certain of these anemias. Indeed, as more delicate techniques are discovered for studying the complexities of the arrangement of the protein constituents of the erythrocyte, it may be expected that eventually all of the members of this group of "hereditary anemias" will be shown to have abnormalities of the hemoglobin or of the supporting stroma.

The types of hereditary anemias are listed in Table I. These will be discussed separately in some detail, especially in regard to pathophysiology, with the exception of ovalocytosis and hereditary nonspherocytic hemolytic anemia, which occur so rarely that relatively little is known concerning the precise hemolytic mechanisms involved. The details of diagnosis and treatment will be discussed by Carl Smith under the title of "Anemias in Infancy and Childhood." Also see Wintrobe's[1] excellent textbook of hematology.

I. CONGENITAL HEMOLYTIC ANEMIA

Mechanism: In congenital hemolytic anemia the abnormal red blood cell manifests itself as a spherocyte, which is inherited as a Mendelian dominant from either parent. A spherocyte is so constructed as to have the largest amount of volume for the smallest amount of surface. All red cells behave as osmometers and take in extra water under circumstances of stagnation such as occur in the spleen. The biconcave shape of the nor-

EFFECT OF SPLEEN ON RED CELLS

Cells	Circulation		Spleen
	Smear	X-Section	X-Section
Normal Erythrocyte			
Spherocyte			

Fig. 1—Congenital hemolytic anemia.

mal erythrocyte is especially adapted to withstand the stress of swelling; whereas the spherocyte, having no expandable surface for its volume, will have to rupture when extra water enters it. Osmotic fragility studies[2] not only demonstrate the increased osmotic fragility of the spherocyte in the peripheral blood, but also show that it is further increased after stasis within the spleen (See Fig. 1). It has also been shown[2,3] that when these patients are transfused with normal red cells the spleen has a preference for trapping the patient's spherocytes rather than the normal cells. This is a second reason for the enlarged spleen being a graveyard for the patient's erythrocytes.

Dr. Castle[2] has emphasized the point that increased osmotic fragility alone will not explain the destruction of erythrocytes in vivo because the body fluids have a tonicity equivalent to that of 0.85 per cent NaCl, and in most cases of congenital hemolytic anemia the hemolysis in hypotonic saline does not begin until 0.75 per cent NaCl or lower. (The hemolysis of normal erythrocytes begins at about 0.44 per cent NaCl.) His group[4] showed that the "*mechanical* fragility" of the spherocyte is also increased, and postulated that in vivo hemolysis is probably the result of the mechanical buffeting of the erythrocyte as it traverses the narrow capillaries at high speed. Splenectomy abolishes the anemia in these patients by removing the organ which increases the osmotic swelling, and therefore the mechanical fragility, of the erythrocyte to the critical bursting point. The final scheme for the mechanism of hemolysis is outlined in Table II.

Although the osmotic and mechanical fragility of the red cells is somewhat reduced by splenectomy, the original spherocytosis is unchanged. This is to be expected since the inherent defect is hereditary and is an intrinsic part of the cell. Electrophoretic studies have failed to

TABLE II—CONGENITAL HEMOLYTIC ANEMIA

1. Spherocytosis (inherited as Mendelian dominant).
2. Increased osmotic fragility and mechanical fragility.
3. Selective sequestration by spleen of spherocytes.
4. *M.F. and O.F. of RBC in spleen increased to critical level.
5. Hemolysis and anemia.
6. Splenectomy is curative by preventing this critical increase of fragility of RBC in the spleen.
7. Original spherocytosis and increase in RBC fragility remain after splenectomy.

* M.F.—mechanical fragility O.F.—osmotic fragility

show the presence of an abnormal hemoglobin.* The exact defect within the erythrocyte is quite unknown, and may even reside in the stroma rather than within the hemoglobin.

Clinical Picture: What are the manifestations of this disease? Although symptoms would be expected to be manifested in early childhood because the spherocytosis is presumably present at birth, for unknown reasons, the disease may not be clinically apparent until early, or even late, adult life. The only symptoms present are usually those referable to anemia. The patient is often jaundiced. Since the anemia may be mild, the aphorism has arisen: "more jaundiced than anemic." The diagnosis is made by the finding of splenomegaly, a hemolytic type of anemia, and spherocytosis. A negative Coombs test and the presence of spherocytosis in one of the parents will further distinguish this disease from the spherocytic hemolytic anemia of the acquired type. In the occasional case where the spherocytosis is minimal and the osmotic fragility is normal, incubation of the blood for eighteen to twenty-four hours will increase the osmotic fragility much more than that of the erythrocytes of a normal person. (Incubation in the test tube is the in vitro equivalent of in vivo stagnation of blood in an enlarged spleen.)

Treatment: The treatment of choice is splenectomy since a clinical cure of the anemia can be expected in all cases.

II. THALASSEMIA MAJOR AND MINOR

Mechanism: The relationship of thalassemia major (Cooley's anemia) to thalassemia minor ("familial microcytic anemia", "target cell anemia")

* Dr. H. C. Lichtman has recently found that ovalocytosis also has no abnormality of hemoglobin by electrophoretic studies using the filter paper method.

was not well defined until the extensive genetic studies of Valentine and Neel[5] in 1944 indicated that the former is the homozygous and the latter the heterozygous manifestation of the anomaly. In other words, inheritance of the abnormal gene from only one parent would result in heterozygosity and therefore in the relatively benign manifestations of thalassemia minor; whereas inheritance of the abnormal gene from both parents would result in homozygosity and therefore in the serious form of the disease, thalassemia major. The great majority of cases have occurred in persons whose ancestry has stemmed from countries bordering the Mediterranean Sea, e.g., in Italians, Greeks, Armenians and Syrians, and presumably indicates genetic connections. However, there are a few reports of Chinese, Indian and Negro patients.

The abnormal erythrocyte is a very small, flat hypochromic cell which appears defective in hemoglobin. The similarities of this microcytic hypochromic anemia to that of iron deficiency anemia are so striking that it seems that there must be some fundamental error in iron metabolism in this disease. This hypothesis is further borne out by the finding of iron deposition in organs, distributed as in hemochromatosis rather than as in hemosiderosis. The flat erythrocyte has a greatly increased resistance to hypotonic saline – (a platycyte being the reverse of a spherocyte)—but it has an increased mechanical fragility and a shortened life span.

In addition to the hemolytic component there seems to be an added component of decreased capacity of blood formation, despite the morphological appearance of normoblastic hyperplasia of the bone marrow. This is shown by the fact that the reticulocytosis is much more inconstant, and of a lesser degree, than in other types of hemolytic anemias. Indeed, it is not uncommon for the reticulocytes to be normal or low even when there are many normoblasts in the peripheral blood. Many patients who are transfused frequently have few remaining circulating cells of their own blood type. In this respect they more closely resemble patients with an erythropoietic arrest such as pernicious anemia than those with "pure" hemolytic anemias, such as sickle cell anemia. It is tempting to postulate that there is a maturation arrest at the level of the basophilic normoblast because of the relative inability to incorporate iron into the hemoglobin molecule. It is interesting that basophilic and polychromatophilic normoblasts are abundant in the marrow, whereas the hemoglobin-containing orthochromatic normoblast is scarce.

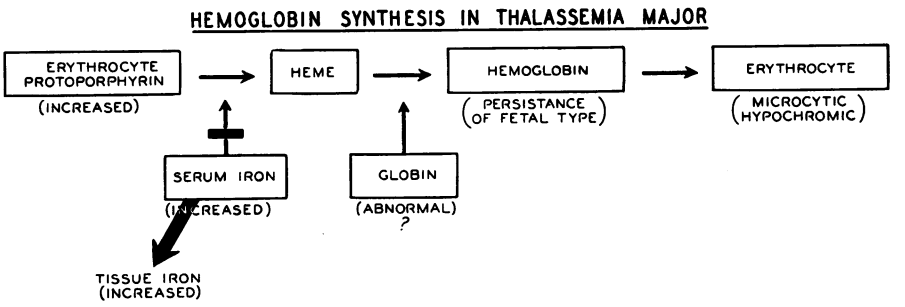

Figure 2.

Recently Singer[6] has shown that there is a persistence of a fetal type of hemoglobin in the red cells in most of these patients, and we have also found that to be so in the course of our studies. Fetal (F) hemoglobin, in contrast to normal (A) hemoglobin, is characterized by a marked resistance to denaturation by alkalis. It has been known that the erythrocytes at birth contain about 80 per cent fetal hemoglobin.[7] This is gradually replaced by normal hemoglobin, so that at the age of four months only about 10 per cent remains, and at the age of one year there is usually none left. The presence of fetal hemoglobin can be demonstrated in both thalassemia major and minor, usually in small amounts, but sometimes in quantities as high as 49 per cent. There is not a good quantitative correlation between the amount of fetal hemoglobin and the severity of the disease. Although the exact significance of fetal hemoglobin is unknown, this is evidence of another abnormality within the hemoglobin itself. Since fetal hemoglobin has been found in a variety of anemias, its presence is thought to be the non-specific result of anemic stress on hemoglobin formation with a partial reversion to fetal pathways. Electrophoretic studies of the hemoglobin of patients with thalassemia have so far not shown any abnormalities in addition to those of a persistence of fetal hemoglobin. A proposed scheme for the mechanism of the anemia in thalassemia is shown in Figures 2 and 3.

Clinical Picture. 1. *Thalassemia major:* This disease is a very serious one with a short life expectancy. The patients develop a severe anemia requiring transfusions by about the second year of life. The marked expansion of the marrow cavity of the malar bones produces the "mongoloid" facies; the generalized marrow changes with their secondary effects on the growing bones produce typical x-ray changes. The spleen

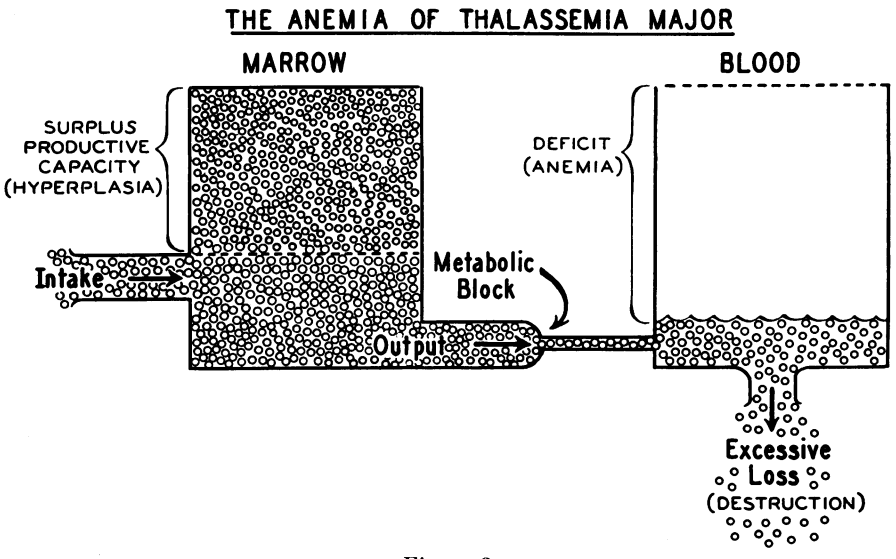

Figure 3.

and liver may become so large as to cause great protuberance of the abdomen. The patient lives an abnormal life at best, having to come to the hospital or clinic for transfusions as often as every two weeks. Death usually intervenes by puberty, as a result of intercurrent infection or of liver or cardiac insufficiency due to hemochromatosis. The diagnosis is easily made in a child of Mediterranean ancestry, who has hepatosplenomegaly, bone changes in the x-rays, and a microcytic hypochromic anemia, with red cells which appear fragmented and which have a marked resistance to hypotonic saline solutions. Occasionally a relatively mild case may simulate the anemia of iron deficiency, but can be differentiated by the lack of response to iron therapy.

The only *treatment* is supportive and consists mainly of blood transfusions. Splenectomy has been shown to ameliorate the anemia in some cases by diminishing the transfusion requirement. Dr. Lichtman[8] has recently found that the favorable effect of splenectomy occurs in patients who have developed an *extracorpuscular* defect as demonstrated by a shortened survival time of normal transfused erythrocytes. This is corrected by splenectomy, thus diminishing the need for blood transfusions. However, since splenectomy does not alter the *intracorpuscular* defect, it is by no means curative, nor has it been ascertained as to whether it lengthens the longevity of the patient.

2. *Thalassemia minor:* This diagnosis is usually based on the results of a chance blood examination of an asymptomatic individual. The findings often consist of a low hemoglobin value and a high red cell count (i.e., a "microcytic polycythemia"), and an increased resistance of the erythrocytes to hypotonic saline. The red cells may have a normal life span. This diagnosis, of course, necessitates no treatment, and the physician should be urged to refrain from producing an "anemic neurosis" in his patient—as is not uncommon! However, there are also many gradations of thalassemia minor, including a shortened red cell survival.[9] These gradations suggest that the thalassemia gene possesses varying degrees of penetrance.

III. Sickle Cell Anemia

Mechanism: In sickle cell anemia the abnormality of the red cell is characterized by its transformation from a biconcave disc to a "sickled" cell when oxygen tension is lowered so that the hemoglobin is in the reduced state. It has long been known that 8 per cent of American Negroes have red cells that can be sickled in vitro and have no anemia or symptoms—i.e., have the "sickle cell trait." About 0.2 per cent have sickle cell anemia, a condition in which the cells can assume the sickled form in vivo as well as in vitro. A basic understanding of the difference between sickle cell anemia and sickle cell trait appeared with Neel's clarification of the hereditary factors[10] and Pauling's demonstration[11] of an abnormal hemoglobin by electrophoretic methods.

Neel showed that sickle cell trait is the expression of heterozygosity of the sickle cell gene and that sickle cell anemia is the expression of homozygosity. For example if there is a mating of two individuals possessing the sickle cell trait, and if they have four children, one child of the four may have sickle cell anemia, two of the four have sickle cell trait, and one may be normal (See Fig. 4). Almost 99 per cent of the cases reported have been in Negroes. The majority of those reported in whites have been Italians or Greeks, and are thought to represent old genetic connections between the peoples of the two shores of the Mediterranean Sea.

Pauling's revolutionary findings published almost simultaneously with Neel's in 1949, showed that the hemoglobin (S) of sickle cells has a different electrophoretic mobility from normal (A) hemoglobin. In sickle cell anemia, all of the hemoglobin is abnormal in this respect;

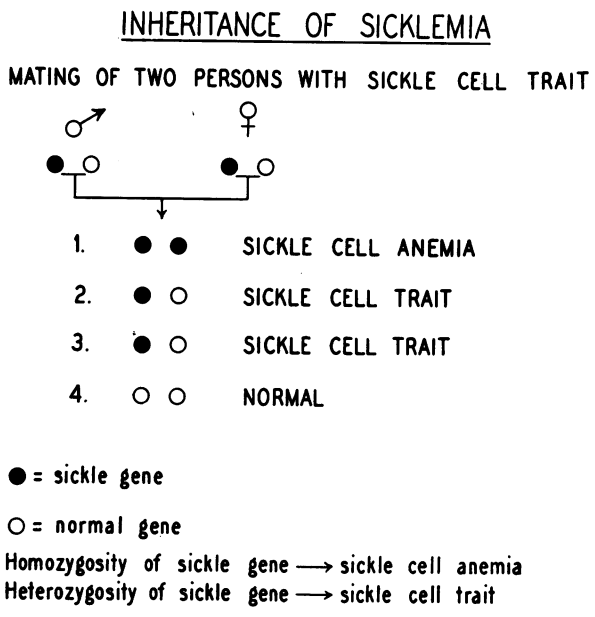

Figure 4.

whereas in sickle cell trait 27-44 per cent of the hemoglobin is S hemoglobin and the remainder of the hemoglobin is normal (A). This obviously fits in very well with the hereditary features just mentioned. The fact that the S hemoglobin is less than 50 per cent may be explained by the hypothesis that the presence of normal hemoglobin in sickle cell trait interferes with complete penetrance of the sickle gene. Electrophoretic studies of heme and denatured globin from S hemoglobin show no abnormalities, but recently studies of natural globins[12] have revealed differences from the normal. X-ray diffraction studies of S and normal hemoglobin crystals are identical. Hence, it is thought that the defect in S hemoglobin may consist in a slightly different arrangement of the basic constituents of the globin portion of the molecule.

Harris[13] has shown that reduced S hemoglobin has a very high viscosity and has a "tactoid" formation which can be observed with the phase microscope. Tactoids are oriented masses of molecules. Perutz[14] has postulated that the change into the sickled shape is the result of crystal formation of S hemoglobin which occurs in the reduced state because of its very low solubility in that form. Reduced S hemoglobin is so insoluble that only 1/7 of it can stay in solution when oxyhemo-

globin changes to reduced hemoglobin. Reduced S hemoglobin was found to have only one-hundredth of the solubility of the oxy-S compound, whereas reduced normal (A) hemoglobin had one-half the solubility of the oxy-A compound. These considerations give us some insight into the dynamics of the extraordinary distortion of the biconcave disc that occurs in the sickling process.

The high viscosity of S hemoglobin in its reduced and relatively insoluble state and the high viscosity which is the inevitable result of the sickle cell *shape* explains one of the worst manifestations of the disease—capillary thrombosis. It is in the capillaries where the O_2 tension falls to its lowest with the production of reduced hemoglobin, so that it is in the capillaries where the sickling takes place with consequent slowing of blood flow due to the increased viscosity. Mass capillary blockade may stop circulation with death of a part, or all, of the organ involved. It is this complication rather than the anemia per se that gives rise to the most distressing symptoms of the disease and eventually to death itself. It has been shown[4] that the mechanical fragility of the cell in the sickled form is many times that of the biconcave disc, so the mechanism of the anemia is also explained by the peculiarity of the sickled shape.

In the case of sickle cell trait there is not enough S hemoglobin in the cell to produce sickling at the lowest O_2 tension (40 mm.) that can occur within the body. It is only in vitro, at artificial levels of much lower O_2 tension (18 mm. or less) that the cells can be changed into the sickle shape. Obviously, under these circumstances no damage can be done in vivo, and the person with sickle cell trait will have no anemia and no symptoms. See Table III for an outline of these mechanisms.

Recently new questions have arisen because studies in Africa have shown that some tribes have an incidence of sickle cell trait of as high as 40 per cent whereas sickle cell anemia is rare or non-existent. This apparent contradiction to the homozygous theory has been explained[15] by postulating that admixture of white "A" genes alters the penetrance of the Negro S gene and thus allows the manifestations of sickle cell anemia! At any rate, much more study is necessary to elucidate these problems. As usual, the solving of one problem opens the door to many more unsolved problems!

Singer[6] has shown that most of his patients with sickle cell anemia have fetal hemoglobin, but that those with sickle cell trait had none.

TABLE III

SICKLE CELL DISEASE

1. RBC will sickle when O_2 tension is reduced.
2. Reduction of O_2 tension in the capillaries.
3. Sickling of RBC.
   a. Increased viscosity → Capillary thrombosis → Tissue necrosis
   b. Increased mechanical fragility → Hemolysis of RBC → Anemia

SICKLE CELL TRAIT

1. RBC will sickle *in vitro* only (very low O_2 tension).
2. RBC will *not* sickle *in vivo*.
3. Therefore no disease.

There was no correlation between the severity of the disease and the amount of the fetal hemoglobin. We have found fetal hemoglobin present in significant amounts in only a few of our patients with sickle cell anemia, the highest value being 39.8 per cent.[16] It is thought that the stress of anemia may cause the organism to resort to old (fetal) methods of producing hemoglobin. However, at birth when there is a high percentage of fetal hemoglobin, infants with sickle cell trait have only a low percentage (about 1-25 per cent) of sickle cells and do not develop 100 per cent of sickle cells until the age of four months, at a time when fetal hemoglobin has almost disappeared. This apparent inability of fetal hemoglobin to sickle may be an important reason for the great rarity of manifestations of clinical sickle cell anemia in the first six months of life.[17]

Clinical Picture: Sickle cell anemia is diagnosed hematologically by the presence of a hemolytic type of anemia, the finding of target cells and a few "irreversible" sickle forms in the stained smear, and 100 per cent sickling in a special preparation in which O_2 tension is reduced by some means (e.g., by sealing with vaseline, reduction with N_2 or H_2 gas, or by reduction with a reducing agent such as sodium bisulphite).

Symptoms are rare in the first six months of life, but are present in half of the cases by the age of two years. Fatigue and irritability occur as a non-specific reflection of the anemia. The more troublesome symp-

TABLE IV—HUMAN HEMOGLOBINS

Type	*F.P. Electro Rate Migration*	*S.C.*	*Incidence*
A (Normal)	Normal	0	99%
F (Fetal)	Slightly Slower	0	Newborn Certain Anemias
S (Sickle)	Much Slower	Pos.	8% Am. Negroes
C	Slowest	0	0.5-2.0% Negroes
D	Same as S	0	?

S, C, or D, if combined with A (heterozygous), are not anemic.
S, C, or D, if combined with each other, are anemic.

F.P. Electro = Filter paper electrophoresis.
S.C. = Sickling.

toms are those which can be attributed to the sickling process itself—i.e., bone and joint pain and abdominal pain and fever. Severe bouts of trouble are known as "crises" and often occur without exacerbation of the hemolytic anemia, and without any known precipitating cause. Many older patients attribute the onset of a crisis to emotional disturbances at home. This points to the vascular factor involved (and may be somewhat analogous to attacks of cardiac decompensation in hypertensives). Death may occur in crisis as a result of insufficiency of any vital organ, such as the brain, lung, liver or kidney. Involvement of the bones leads to typical x-ray lesions[18] and may cause severe pain. Intractable leg ulcers may follow slight trauma and are presumably due to the poor circulation in those areas, complicated by the sluggish flow of sickled cells. Although the spleen often becomes abnormally small because of auto-infarction from sickling within the spleen, this organ may also be enlarged and even give rise to a superadded factor of "hypersplenism."[19]

Treatment: There is no satisfactory treatment, other than supportive, at present. Transfusions should be given for symptoms due to anemia only. The administration of nasal oxygen does not result in clinical improvement and may actually intensify the anemia as a result of the suppression of blood formation. Cortisone and ACTH have been tried with quite variable results, and are of limited value at best. The same can be said for vasodilators.

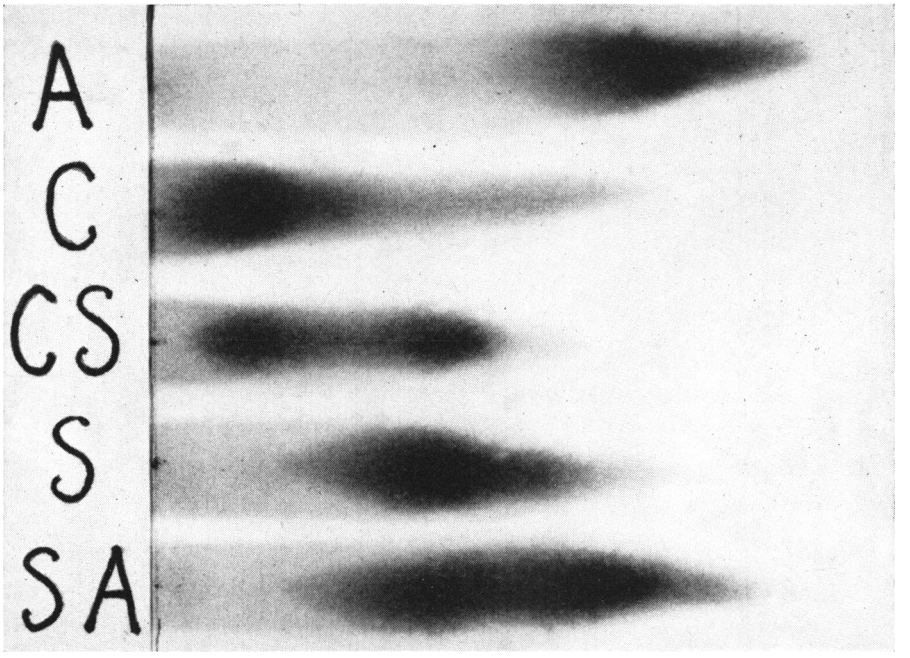

Fig. 5—Human hemoglobins. Filter paper electrophoresis.

For a recent excellent detailed review of sickle cell anemia, the reader is referred to Margolies.[20]

IV. Other Anemias with Abnormal Hemoglobins

In the course of doing genetic studies of the families of Negro patients with sickle cell disease two other hemoglobins, designated C and D,[21] have been discovered[12] (See Table IV). Neither has the ability to produce sickling. Hemoglobin D possesses the same electrophoretic mobility as S hemoglobin, but has the same solubility as normal A hemoglobin. (Reduced S hemoglobin has only 1/14 the solubility of A hemoglobin in the reduced state.) Hemoglobin C in filter paper electrophoresis migrates very slowly, much slower than S hemoglobin, which in turn migrates more slowly than normal hemoglobin (See Fig. 5).

Any combination of these abnormal hemoglobins other than with normal hemoglobin will result in a hemolytic anemia (See Table V). If the abnormal hemoglobin is heterozygous with normal hemoglobin, an asymptomatic and non-anemic trait will be present. These abnormal

TABLE V—DISEASES ASSOCIATED WITH ABNORMAL HEMOGLOBINS

Disease	*HGB.*	*Anemia*	*Targets*	*Crises*	*Splenomegaly*
Thal.	A (& F)	Microcytic	1+	0	4+
S.C.A.	S-S (& F)	Normocytic	2+	4+	±
S.C.-Thal.	S-A (& F)	Microcytic	3+	1+	2+
HGB. C	C-C	Normocytic	4+	0	0
HGB. D	D-D	?	?	?	?
S.C.-C	S-C	Normocytic	3+	1+	2+
S.C.-D	S-D	?	?	?	?

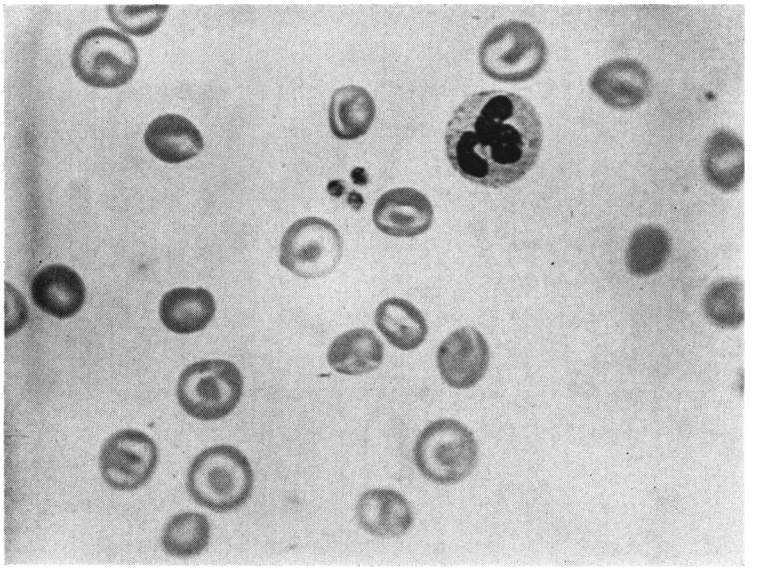

Fig. 6—Hemoglobin C disease. Target cells.

hemoglobins depend on a single Mendelian factor, and have been found in Negroes but not in whites.[12, 22, 23]

The Hereditary Anemias. 1. *Hemoglobin C Disease:* Although three observers have seen examples of homozygous hemoglobin C, the reports are not yet published. Our one case[24] is characterized by mild anemia (hemoglobin 11 gm. per cent) and 60-90 per cent target cells in the peripheral blood (See Fig. 6). The patient has mild arthralgia and abdominal pain, but no crises. The spleen is not palpable. The exact mechanism that leads to the destruction of the red cells is not at all

clear, other than the fact that target cells are apparently mechanically fragile. Why the abnormal hemoglobin is associated with target cell formation is another unanswered question.

2. *Hemoglobin C–Sickle Cell Disease:* The combination of hemoglobin C and S[22, 23] occurs in a ratio of 1:1 and results in a mild hemolytic anemia with mild crises, splenomegaly and a great increase in target cells. The incidence of hemoglobin C, occurring as a heterozygous trait, or in combination with C or S, has been variously reported to occur in 0.5 per cent to 2 per cent of American Negroes.[22, 23] About 30-40 per cent of the hemoglobin of individuals heterozygous for this factor is electrophoretically abnormal.

3. *Hemoglobin D Disease:* Homozygous hemoglobin D has not yet been found, but its presence can be predicted. Only two instances of hemoglobin D trait (heterozygous) have so far been reported,[25] and these were in two Caucasian siblings. Clinical and hematologic data have not yet been given.

4. *Thalassemia–Sickle Cell Disease:* The combination of the thalassemia gene with the sickle gene apparently favors penetrance of the sickling gene, since about 70 per cent of the hemoglobin is S hemoglobin. About nineteen families have been studied where this combination was found.[26] The clinical picture is similar to that of relatively mild sickle cell anemia, but the spleen is usually enlarged. Hematologically the anemia is hemolytic in type, but is microcytic and hypochromic with many target cells.

To summarize, if a patient, who *appears* to have sickle cell anemia, is found to have only *one* parent who has the sickling trait, investigation of the non-sickling parent (if he *is* the true parent) should show that he has either 1) hemoglobin C, 2) hemoglobin D, 3) thalassemia, or 4) an anomalous form of sickle cell trait,[27] in which less than 10 per cent of the hemoglobin is of the S type. Future investigation may also add to this list of abnormal hemoglobins and their combinations (such as "Thalassemia-Hemoglobin C Disease," not yet reported). Some of the atypical hemolytic anemias of the hereditary type, undiagnosed at present, may be clarified by future studies along analogous lines.

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